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| SERIAL NUMBER | FILING DATE | FIRST NAMED INVENTOR | ATTORNEY DOCKET NO. |
|---|---|---|---|
| 08/327,522 | 10/21/94 | LOCKHART | D 16528X86 |

| EXAMINER | |
|---|---|
| FREDMAN, J | |
| **ART UNIT** | **PAPER NUMBER** |
| 1807 | 5 |

18N1/0807

EUGENIA GARRETT-WACKOWSKI
TOWNSEND AND TOWNSEND KHOURIE AND CREW
ONE MARKET PLAZA 20TH FLOOR
STEUART STREET TOWER
SAN FRANCISCO CA 94105

**DATE MAILED:** 08/07/95

This is a communication from the examiner in charge of your application.
COMMISSIONER OF PATENTS AND TRADEMARKS

☑ This application has been examined ☑ Responsive to communication filed on 2-27-95 ☐ This action is made final.

A shortened statutory period for response to this action is set to expire - 3 - month(s), - 0 - days from the date of this letter.
Failure to respond within the period for response will cause the application to become abandoned. 35 U.S.C. 133

**Part I THE FOLLOWING ATTACHMENT(S) ARE PART OF THIS ACTION:**

1. ☑ Notice of References Cited by Examiner, PTO-892.
2. ☑ Notice of Draftsman's Patent Drawing Review, PTO-948.
3. ☐ Notice of Art Cited by Applicant, PTO-1449.
4. ☐ Notice of Informal Patent Application, PTO-152.
5. ☑ Information on How to Effect Drawing Changes, PTO-1474.
6. ☐ _____.

**Part II SUMMARY OF ACTION**

1. ☑ Claims 1-18 are pending in the application.

   Of the above, claims _____ are withdrawn from consideration.

2. ☐ Claims _____ have been cancelled.

3. ☐ Claims _____ are allowed.

4. ☑ Claims 1-18 are rejected.

5. ☐ Claims _____ are objected to.

6. ☐ Claims _____ are subject to restriction or election requirement.

7. ☑ This application has been filed with informal drawings under 37 C.F.R. 1.85 which are acceptable for examination purposes.

8. ☐ Formal drawings are required in response to this Office action.

9. ☐ The corrected or substitute drawings have been received on _____. Under 37 C.F.R. 1.84 these drawings are ☐ acceptable; ☐ not acceptable (see explanation or Notice of Draftsman's Patent Drawing Review, PTO-948).

10. ☐ The proposed additional or substitute sheet(s) of drawings, filed on _____. has (have) been ☐ approved by the examiner; ☐ disapproved by the examiner (see explanation).

11. ☐ The proposed drawing correction, filed _____, has been ☐ approved; ☐ disapproved (see explanation).

12. ☐ Acknowledgement is made of the claim for priority under 35 U.S.C. 119. The certified copy has ☐ been received ☐ not been received ☐ been filed in parent application, serial no. _____; filed on _____.

13. ☐ Since this application apppears to be in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under Ex parte Quayle, 1935 C.D. 11; 453 O.G. 213.

14. ☐ Other

# 08/327,522
**EXAMINER'S ACTION**

PTOL-326 (Rev. 2/93)

**Part III DETAILED ACTION**

***Sequence Rules***

1. This application complies with the Sequence Rules and the sequences were entered by the Scientific and Technical Information Center.

***Drawings***

2. The Office will accept black and white photographs in utility and design patents only after granting a petition filed under 37 CFR 1.84 which requests that photographs be accepted. The petition must be granted before the photographs can be approved by the Official Draftsman. The petition must include the appropriate fee set forth in 1.17(h) and three sets of photographs.

***Specification***

3. The specification is replete with spelling errors too numerous to mention specifically. The specification should be revised carefully. Examples of such errors are: On page 4, line 3, "nucelic" should be --nucleic--; on page 4, line 4, "labelled" should be --labeled--; on page 4, line 7, "oligonucelotide" should be --oligonucleotide--.

***Claim Rejections - 35 USC § 112***

4. The following is a quotation of the first paragraph of 35 U.S.C. § 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains,

or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-18 are rejected under 35 U.S.C. § 112, first paragraph, as the disclosure is enabling only for claims limited to target nucleic acids shorter than 22 nucleotides in length of previously defined sequence. See M.P.E.P. §§ 706.03(n) and 706.03(z). While the breadth of these claims includes target nucleic acid of any length and known or unknown sequence the specification lacks guidance on a variety of information necessary to determine unknown sequences or long sequences. Specifically, no algorithms by which the data would be analyzed are presented. The absence of information regarding data analysis is critical, since it does not appear to be a simple problem to analyze varying intensity signals from a variety of oligonucleotides, and correctly identify the correct sequence even with the addition of enzymatic discrimination. Further, unknown sequences may pose a variety of problems for the technique and software. Unknown sequences with long repeats, greater than the length of the interpreting oligos will be unreadable by this technique. The use of enzymatic discrimination adds another set of sequences which will be uninterpretable, which are sequences which are of equal length to the oligonucleotide but differ in a single base pair. Such sequences would previously have been read as ambiguous, but now may allow for correct identification of a first sequence at the

expense of an inability to identify or incorrectly identify a second sequence. The amount of direction or guidance presented in the specification is minimal given that no information regarding modes of data analysis, duplicate sequences which differ by a single base pair, or repeat sequences is present. There are no working examples of sequencing initially unknown sequences, or sequences containing duplications, nor sequences of sufficient complexity to truly require computer assisted analysis. There is some prior art (Drmanac et al U.S. Patent 5,202,231) which discusses sequencing by hybridization and the problem of repeat sequences but does not give a solution which will allow for sequencing of any repeats. Although the level of skill in the art of nucleic acid hybridization is high (the Ph.D. degree with laboratory experience), there is no predictability for which sequences would be capable of being sequenced in this method, nor is there any predictability for which mode of data analysis is capable of discriminating the exceedingly complex pattern that would result from a sequence of ordinary genomic length. A gene of only 2000 nucleotides would consist of 1992 8-mers, some of which might represent duplicates, and would represent approximately 3% of all 8-mer sequences, or 48% of all 6-mer sequences. If the gene was the size of the b-globin locus, which exceeds 60,000 basepairs in size and would have more than 60,000 6- or 8-mers, it could potentially utilize every possible 6- or 8-mer sequence. It is unclear how such a gene would be

analyzed by the method disclosed, particularly using the arrays of claims 5-7 which are sufficiently small to be overwhelmed by relatively short sequences. The quantity of experimentation that would be necessary to determine methods by which any sequence of any size could be sequenced versus the specific 22 mers shown is substantial. Accordingly, undue experimentation is required to make and use the invention as broadly claimed.

5. Claims 1-18 are rejected under 35 U.S.C. § 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. It is vague and indefinite what is meant by the term "positionally distinguishable oligonucleotides each of which binds to a defined subsequence of preselected length" in claims 1 and 17. It is unclear whether these oligonucleotides are oligonucleotides of varying sequence, combined together, and located in a single location, with other oligonucleotides of varying sequence, combined together, located in other locations or whether a single oligonucleotide sequence is located in each location. It is vague and indefinite what are the metes and bounds of "substantially" in claim 2. It is unclear what constitutes "substantially all possible subsequences". It is vague and indefinite what are the metes and bounds of claims 3 and 4 since the modifier "about" is used in connection with a range. While "about" a specific number indicates a reasonable (such as 10%) variability is acceptable,

"about" a range carries an unclear meaning. It is vague and indefinite what is meant by the term "unligatable" in claim 17, step (c). It is unclear if this is simply a typographical error in which "unligatable" should have been --unligated-- or if some other meaning which is unclear is meant.

Also, method claims require a last step or phrase in the last step that states the accomplishment of the goals for the method which were stated in the method's preamble. Claims 1 and 17 lack such a last step and are confusing because the additional method step is not sufficiently set forth. While minute details are not required in method claims, at least the basic steps must be recited in a positive, active fashion. See Ex parte Erlich, 3 USPQ2d1011, p.1011 (Bd. Pat. App. Int. 1986). It is suggested that an amended claim more clearly describing the intended conclusion step(s) be submitted.

***Claim Rejections - 35 USC § 103***

6. The following is a quotation of 35 U.S.C. § 103 which forms the basis for all obviousness rejections set forth in this Office action:

> A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.
>
> Subject matter developed by another person, which qualifies as prior art only under subsection (f) or (g) of section 102 of this title, shall not preclude patentability under this section where the subject matter and the claimed invention

were, at the time the invention was made, owned by the same person or subject to an obligation of assignment to the same person.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. § 103, the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 C.F.R. § 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of potential 35 U.S.C. § 102(f) or (g) prior art under 35 U.S.C. § 103.

7. Claims 1-16 are rejected under 35 U.S.C. § 103 as being unpatentable over Fodor et al (WO 92/10588) in view of Sambrook et al (Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, p. 5.80, 7.58-7.78)

Claim 1 is drawn to a method of sequencing comprising the steps: a) combining $a_i$) an oligonucleotide array, $a_{ii}$) a target nucleic acid to form hybrid complexes, b) adding a nuclease to digest hybrid complexes which are not perfectly complementary, c) detecting remaining complexes bound to oligonucleotides. Claim 2 limits claim 1 to an array which can detect all sequences. Claim 3 limits claim 1 to oligonucleotides between 6 and 20 bases. Claim 4 limits claim 1 to oligonucleotides between 8 and 15 bases. Claim 5 limits the array to $10^3$ different oligos. Claim 5 limits the array to $3 \times 10^3$ different oligos. Claim 7 limits the array to $10^4$ different oligos. Claim 8 limits the array to $10^5$ different oligos. Claim 9 limits the array to $10^6$ different oligos. Claim 10 limits claim 1 to a target nucleic acid which is RNA. Claim 11 limits claim 10 to an RNA nuclease. Claim 12

limits claim 11 to RNAse A. Claim 13 limits claim 1 to a target nucleic acid which is DNA. Claim 14 limits claim 13 to a DNA nuclease. Claim 15 limits claim 14 to S1 nuclease. Claim 16 limits claim 14 to Mung bean nuclease. Claim 17 is an embodiment of claim 1 in which a ligation step of adding a detectable probe to the hybrid complexes is performed in the place of nuclease treatment. Claim 18 limits claim 17 to the use of T4 DNA ligase.

Fodor teaches a method of sequencing comprising the steps: a) combining $a_i$) an oligonucleotide array composed of 8- to 15-mers (page 26) and up to $10^6$ oligonucleotides (page 11), $a_{ii}$) a target nucleic acid to form hybrid complexes, c) detecting remaining complexes bound to oligonucleotides (page 4, also pages 34-36).

Fodor does not teach step b) adding a nuclease to digest hybrid complexes which are not perfectly complementary nor the use of specific nucleases.

Maniatis teaches methods of S1 mapping and ribonuclease protection assays in which S1 nuclease or RNAse A are used to remove unbound and imperfectly complementary hybrid complexes (pages 7.58-7.78). Maniatis also teaches that Mung Bean nuclease is functionally equivalent to S1 nuclease (page 5.80).

It would have been *prima facie* obvious to one having ordinary skill in the art at the time the invention was made to combine the sequencing method of Fodor with the use of nuclease to distinguish perfectly complementary hybrid complexes from

imperfectly complementary complexes as taught by Maniatis since Maniatis states that in S1 mapping "DNA that has not formed duplexes is hydrolyzed with nuclease S1, whereas DNA that has hybridized to RNA is protected from digestion (page 7.58)". Maniatis further states in regard to ribonuclease protection assays "The sensitivity of this method is therefore approximately 20-fold greater than that attained with double-stranded DNA probes . . . Furthermore, the digestion of RNA:RNA hybrids with RNAase appears to suffer fewer artifacts than digestion of RNA:DNA hybrids with nuclease S1. For these reasons and because of the relative ease with which radiolabeled RNA probes can be synthesized, it is not surprising that RNAase digestion of RNA:RNA hybrids has become a standard method to quantitate mRNA molecules (page 7.71, paragraph 3)". An ordinary practitioner would have been motivated to combine the method of Fodor with the nuclease methods of Maniatis for the explictly stated benefits of sensitivity, reduction of artifacts, and ease of use in the removal of imperfectly hybridizing molecules for better detection of perfectly hybridizing molecules.

8. Claims 17 and 18 are rejected under 35 U.S.C. § 103 as being unpatentable over Fodor in view of Keith et al (U.S. Patent 5,093,245).

Claim 17 is drawn to a method of sequencing comprising the steps: a)combining $a_i$) an oligonucleotide array, $a_{ii}$)a target nucleic acid to form hybrid complexes, b) ligating a labelled

oligonucleotide probe to the hybrid complex, c) removing the unligated probed, and d) detecting remaining complexes bound to oligonucleotides. Claim 18 limits claim 17 to the use of T4 DNA ligase.

Fodor teaches a method of sequencing comprising the steps: a) combining $a_i$) an oligonucleotide array composed of 8- to 15-mers (page 26) and up to $10^6$ oligonucleotides (page 11), $a_{ii}$) a target nucleic acid to form hybrid complexes, c) detecting remaining complexes bound to oligonucleotides (page 4, also pages 34-36).

Fodor does not teach the ligation of labeled oligonucleotides for detection of DNA, nor does Fodor teach the use of T4 DNA ligase in that detection.

Keith teaches the ligation of labeled oligonucleotides with T4 DNA ligase for the detection of DNA (column 2, lines 29-42 and columns 7 and 8).

It would have been *prima facie* obvious to one having ordinary skill in the art at the time the invention was made to combine the sequencing method of Fodor with the ligation of labeled oligonucleotides method of Keith since Keith states "It is evident from the above results that a simple effective process for labeling or modifying termini of double-stranded DNA is provided. Smaller amounts of the labeling moiety are required, whil oligomerization of the sample is substantially prevented. Labeling can occur in the same reaction vessel in which

restriction or specific fragmentation is accomplished. Thus a homogenous product is obtained which provides for accurate sizing, detection and ease of further manipulation (column 9, lines 3-11)". An ordinary practitioner would have been motivated to combine the methods of Fodor and Keith for the expressly stated and expected benefits of simplicity, accuracy of detection, and ability to use small amounts of the label.

9. No claims are allowable over the prior art.

***Conclusion***

10. The prior art made of record and not relied upon is considered pertinent to applicant's disclosure. Drmanac et al (U.S. Patent 5,202,231) Sequencing by hybridization. Pirrung et al (U.S. Patent 5,143,854) Lithography of polypeptides. Fino (U.S. Patent 5,290,925) Ligation of oligonucleotides. Southern (WO 89/10977) Sequencing by hybridization. Hod (Biotechniques (1992) 13(6):852,854) Ribonuclease protection assay.

11. No claims are allowed.

12. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Jeff Fredman, Ph.D. whose telephone number is (703) 308-6568.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, W. Gary Jones, can be reached on (703) 308-1152.

Any inquiry of a general nature or relating to the status of this application should be directed to the Group receptionist whose telephone number is (703) 308-0196.

Papers related to this application may be submitted to Group 180 by fascimile transmission via the P.T.O. Fax Center located in Crystal Mall 1. The CM1 Fax Center number is (703) 308-7939. Please note that the faxing of such papers must conform with the Notice to Comply published in the Official Gazette, 1096 OG 30 (November 15, 1989).

Jeffrey Fredman, Ph.D.

August 2, 1995

W. GARY JONES
SUPERVISORY PATENT EXAMINER
GROUP 1800

8/3/95